

REMARKS

This is in full and timely response to the above-identified Office Action. The above listing of the claims supersedes any previous listing. Favorable reexamination and reconsideration are respectfully requested in view of the preceding amendments and the following remarks.

Rejections under 35 USC § 112

In this response, claim 4 has been amended in a manner which is submitted as rendering the claimed subject matter both clear and concise. More specifically, this claim has been amended to overcome the objection that limitation "at most 10 times, preferable at most 5 times, in particular at most 2 times" is considered unclear.

In particular, it has been stated that it is unclear whether the limitation applies to each of the extension reactions or only to a specific extension reaction. In this regard, please refer to section [0038] of the present application from which it becomes clear that the limitation may apply to either of the extension steps. Claim 4 has been amended to read:

"The method as claimed in claim 1, wherein the first primary extension reaction is carried out under the first conditions at most 10 times, preferably at most 5 times and in particular at most 2 times and/or the second primary extension reaction is carried out under the second conditions at most 10 times, preferably at most 5 times and in particular at most 2 times".

Dependent claims 23 and 25 were objected to in that these claims refer to an electrode which finds no antecedent basis in claim 1. This issue is resolved by the amendments which render these claims dependent on claim 22 which provides the requisite antecedent basis.

Rejections under 35 USC §103

The rejection of claims 1-2, 4-9, 12-19 and 21 under 35 USC § 103(a) as being unpatentable over Wong et al. (US patent 5,935,793) in view of the article to Heath et al. is respectfully traversed.

Applicant notes that the Examiner asserts that Wong et al. teaches a method comprising essentially similar the steps as those recited in present claim 1. Applicants respectfully traverse this conclusion. Wong et al. describes a completely different method and is directed to the parallel sequencing of a group of different target polynucleotides.

The different purpose of the method described by Wong as compared to the present application is reflected by the fact that some important method steps referred of present claim 1 are clearly not taught by Wong.

First, as also acknowledged by the Examiner at page 10 of the office action, Wong does not teach the use of a first PCR primer pair as required in step a) of present claim 1. In contrast, Wong only teaches the use of a single primer to be employed for primer extension reaction, thereby generating "sequencing fragments" by the well-known chain termination method of Sanger. In this method, a primer extension reaction is carried out in the presence of a particular dideoxynucleotide (ddATP, ddGTP, ddCTP, or ddTTP). Incorporation of a dideoxynucleotide into the DNA strand synthesized in the primer

extension reaction therefore terminates DNA strand extension and results in DNA fragments of varying length (the "sequencing fragments"). The sequencing fragments are then separated on the basis of their lengths under conditions effective to resolve fragments differing in length by a single base in order to produce a plurality of resolved size-separated bands. The size-separated bands are then collected in separate aliquots for further amplification of the identifier tag sequence.

According to the first step of the claimed method, the primer containing the tag identifier sequence is used in a PCR reaction for amplifying target nucleic acid which is flanked by primers P1 and P2 of the first primer pair. Thus, step a) of the method as presently claimed is not taught by Wong. Plainly, Wong does not provide a second primer which would correspond to the P1 primer of the present invention.

Furthermore, it can be taken from the language of step a) of present claim 1 that the second primer P2 exhibits a 5'-terminal segment (c3) and a 3'-terminal segment (c4) which may be spaced from each other by the tag segment (designated i). This general assembly is reflected in particular in figure 1 of the present application. It is required that segment c4 of primer P2 can specifically hybridize with a predetermined segment of the nucleic acid to be detected. That is to say, the c4 portion of primer P2 is specific for each of the nucleic acid molecules in the sample. In contrast, the primer used in the initial extension step of Wong (which is apparently considered by the Examiner to correspond to primer P2) hybridizes to the sequence of the cloning vector and not to any of the sample fragments used in the method of Wong. Accordingly, this feature of present claim 1 is also not reflected by the method of Wong.

Likewise, there is no step in the method of Wong which could be regarded as an extension of primers P1 and P2 of the first primer pair wherein the extension products resulting from the extension of the P1 and P2 primer themselves serve as a template for binding of P1 and P2 primers for generating additional extension products. This is of course not surprising, since the method of Wong does aim at the amplification of the target nucleic acid, but to the generation of dideoxynucleotide-terminated fragments to be used in downstream sequencing steps. Consequently, step c) of method claim 1 is not fulfilled in the method of Wong.

Finally, as outlined in step d) of method of claim 1, a PCR is carried out with a third and fourth primer (P3 and P4, respectively) which amplifies the complete portion of the nucleic acid which is flanked by primers P3 and P4.

In contrast, Wong only teaches amplification of the tag identifier within the primer-tag-primer structure depicted in figure 1B. It is explicitly stated in column 10, line 15 onwards that the primer-tag-primer shown in figure 1B has the advantage that it allows rapid exponential amplification of the tag identifier in each sequencing fragment without amplifying the sample fragment sequences. In other words, Wong clearly teaches away from amplifying anything more than the tag identifier. Consequently, step d) of method claim 1 is not fulfilled in the method of Wong.

In light of the above, it is directly apparent that the method disclosed by Wong can under no circumstances be compared to the method referred to in claim 1 of the present application. The present application relates to a method which enables PCR-based detection of different nucleic acids in parallel. A person skilled in the art who faces to solve this problem would not

have taken into consideration the disclosure of Wong, since it relates to a remote technical field and the skilled person would not have expected to find any helpful hints in such publication which would assist him in solving the problem underlying the present invention.

At page 10 of the office action, the Examiner states that Wong does not explicitly teach the use of a PCR primer pair in step a) and instead refers to the first step of the method as requiring at least one primer for the extension of the original sample sequences. Here, the Examiner apparently intends to imply that the method according to Wong could also be carried out with a PCR step using a primer pair as disclosed in step a) of claim 1 of the present application. This is certainly not a convincing argument.

Due to the different purposes for which the method of Wong on the one hand and the method of the present application on the other hand are used, it would simply make no sense to carry out the primer extension reaction referred to in Wong with two primers. Clearly, the primer extension reaction referred to in Wong is a Sanger reaction which shall provide for the generation of different fragments with different lengths and a predetermined terminal dideoxynucleotide. Applying the PCR reaction with two primers at this stage of the Wong method would completely contravene the purpose of that method.

The subject matter of claim 1 is also not rendered obvious by a combination of Wong and Heath (Journal of Medical Genetics, 2000, 37:272-280).

Heath describes a multiplex PCR approach using universal primers. Heath completely lacks any indication of using any kind of tag as an identifier of a particular PCR product. It should be noted in this context that the term "tag" as used in the

disclosure of Heath does not refer to a specific polynucleotide sequence which is used as an identifier. It simply refers to the fact that one specific primer sequence in a primer molecule may be tagged with another (universal) primer sequence. Accordingly, the method of Heath does not describe the use of immobilized probes which specifically hybridize with tag identifier of the amplification products.

It is unclear as to why the hypothetical person of ordinary skill would consider combining the teaching of Wong with those of Heath. Both methods serve completely different purposes. It is to be noted that such combination of documents appears rather artificial, since Wong refers to a method of sequencing polynucleotides, whereas Heath describes a multiplex PCR approach for detecting gene rearrangements.

From a technical point of view, the only feature shared by the methods of Heath and Wong resides in the use of universal primers at some stage of the process. However, this can certainly not be regarded as a sufficient reason to combine the teachings of these in manner that might arrive at the subject-matter of claim 1 of the present application.

Conclusion

In conclusion, none of the publications of Heath or Wong is able to render obvious the subject-matter of present claim 1. Therefore, claim 1 as presently pending is clearly non-obvious in light of the art which is applied. Dependent claims 2-37 are non-obvious and allowable over the cited art for at least the same reasons that claim 1 is so allowable.

It is respectfully submitted that the claims as they have been amended are therefore allowable over the art which has been

applied in this Office Action. Favorable reconsideration and allowance of this application are courteously solicited.

Three month extension of time is hereby requested. A credit card authorization form in the amount of \$1,050.00 is attached herewith for the three month extension of time.

Respectfully submitted,

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